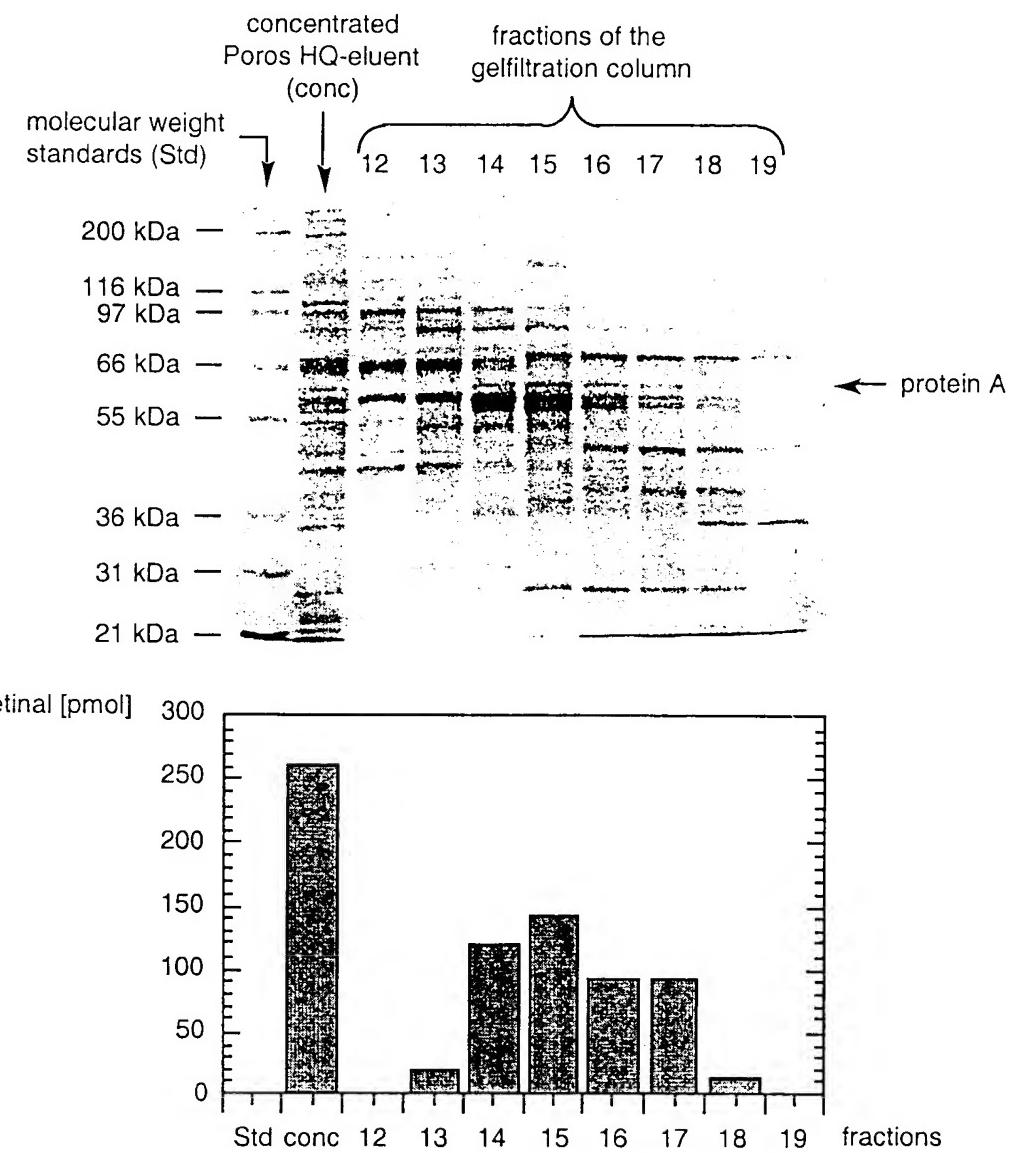


Figure 1



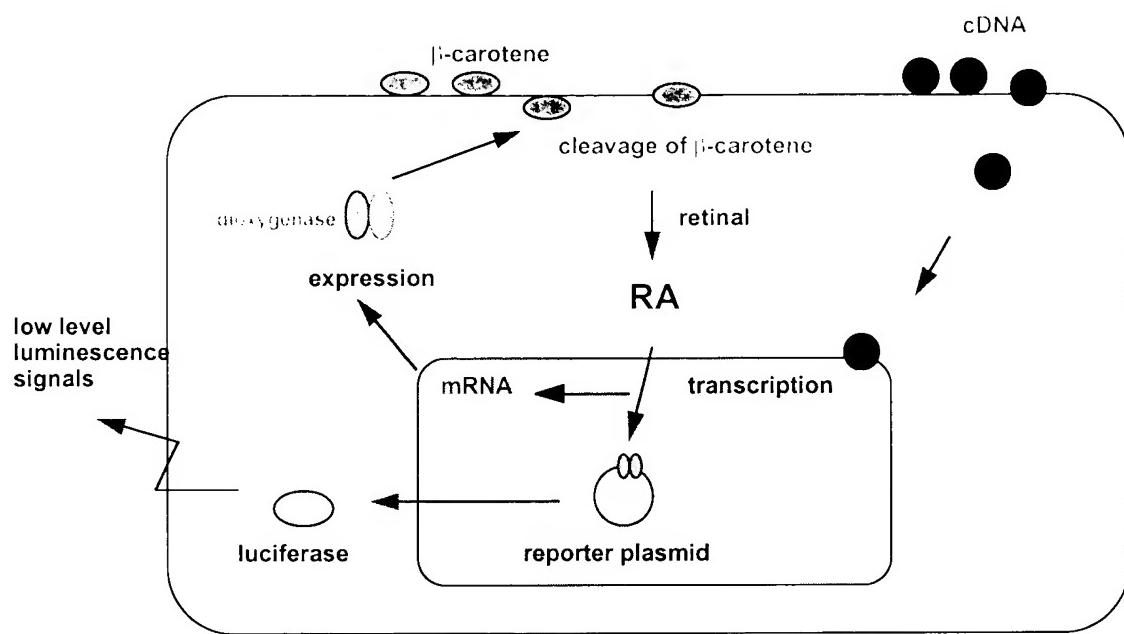


Figure 2

Figure 3  
No. 2

Seq. ID

1 CGGATCCACT AGTAACGGCC GCCAGTGTGG TGGAATCCAT  
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51 AACAGGAAAG AGCTGTTCTT AGGCCAGAGA GGAGGGCACC  
GTACGCCCTGC

101 AGGAGCAGCT GGGTAGAGGA CACAGGAGAG CGATGGAGAC  
AATATTTAAC

151 AGAAACAAAG AAGAGCATCC AGAGCCCATA AAAGCTGAGG  
TGCAAGGTCA

201 GTTGCCTCACT TGGTTGCAAG GGGTACTTCT CCGAAATGGC  
CCAGGGATGC

251 ACACAATAGG GGACACTAAA TACAACCCT GGTTTGATGG  
CTTGGCTCTG

301 CTGCACAGCT TCACGTTAA AAATGGTCAA GTTTACTACA  
GAAGTAAGTA

351 CCTCCGAAGT GACACATACA ACTGCAATAT AGAAGCAAAC  
CGAATCGTGG

401 TGTCTGAGTT TGGAACCATG GCTTATCCGG ATCCATGCAA  
AACATATTT

451 GCCAAGGCAT TCTCATACTT ATCTCACACC ATTCCCTGAGT  
TCACGGACAA

501 CTGCCTGATC AACATTATGA AAACCTGGGA TGATTATTAT  
GCTACCAGTG

551 AGACTAACTT CATCAGAAAA ATTGATCCAC AGACTCTGGA  
GACACTAGAT

601 AAGGTAGACT ACAGCAAATA TGTAGCTGTA AACTTGGCAA  
CTTCTCACCC

651 ACACATGAC AGTGCTGGAA ATATTCTCAA CATGGGTACT  
TCAATTGTTG

701 ATAAAGGGAG AACAAAATAT GTTCTCTTA AGATCCCTTC  
CTCTGTACCA

751 GAAAAAGAAA AGAAGAAATC TTGTTTTAAA CAACCTGGAAG  
TAGTATGCTC

801 CATCCCTTCT CGCTCCCTGC TCCAACCAAG CTACTACCAC  
AGCTTGGAA

851 TCACAGAAAA TTATATTGTG TTCATAGAGC AGCCATTAA  
ACTGGATATT

901 GTCAAACCTGG CAACTGCCTA CATCCGAGGT GTGAACCTGGG  
CTTCCTGCCT

951 TTCCTTTCAT AAGGAGGATA AGACGTGGTT TCACCTTGTA  
GACAGAAAGA

1001 CGAAAAAAAGA AGTATCCACC AAGTTTTACA CTGATGCTTT  
GGTGCTTTAT

1051 CACCACATAA ATGCTTACGA AGAAGATGGC CACGTTGTTT  
TTGATATCGT

1101 TGCCTACAGA GACAATAGCT TGTACGATAT GTTTTACTTA  
AAAAAAACTGG

1151 ACAAAAGACTT TGAAGTGAAC AACAAAGCTTA CCTCCATCCC  
AACCTGCAAG

1201 CGCTTGTG TGCGCTCTGCA GTATGACAAG GATGCAGAAG  
TAGGTTCTAA

1251 TTTAGTCAAA CTTCCAACCTT CCGCAACTGC TGTAAAAGAA  
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1301 GCATCTATTG TCAACCTGAA ATATTATGTG AAGGGATAGA  
ACTGCCTCGT

1351 GTCAACTATG ACTACAATGG CAAAAAAATAC AAGTATGTCT  
ATGCAACAGA

1401 AGTCCAGTGG AGCCCAGTTC CTACAAAGAT TGCAAAACTG  
AATGTCCAAA

1451 CAAAGGAAGT ACTGCACTGG GGAGAAGACC ACTGCTGGCC  
CTCAGAGCCC

1501 ATCTTGTTC CCAGCCCCGA TGCAAGAGAA GAGGATGAAG  
GTGTTGTTT

1551 GACCTGTGTT GTGGTGTCTG AGCCAAATAA AGCACCCCTTC  
CTACTCATCT

1601 TGGATGCTAA AACATTCAA GAATTGGGCC GAGCCACAGT  
TAACGTAGAA

1651 ATGCATCTGG ACCTGCATGG GATGTTTATA CCACAGAATG  
ATTTGGGGC

1701 TGAGACGGAA TAAAACGCTA TTGATCCGAC TACACAAACT  
GAGACAAACTT

1751 TCTACTGAAC ATGAGTTAAT ATCCCTTTA CCATTCAAGA  
ACAACCATAT

1801 AACGACACAA AATGACTATG TATAATCTCT TAAATAATAG  
ATATAATCCT

1851 TTTAAGGCAC AGCGATGAGT TTTACTACAG GTAACGATAT  
GCACAACTGG

1901 CATATAACTA TTCCAAAAGA AGAAGAACGA TCAGTGT  
AGAAGTGCTA

1951 ATGTTGTACA TAACGGCGGC AGAGGGAAACA GGAGAGAAAG  
GTAACGGGAA

2001 TATTTAATAG AATATAGATT TCTGAGCAAA TGAAGTGCAG  
TATTATGGT

2051 GTGATGCATG GCATGAGTCA CATAGGTCTG CAGCTCATGT  
ATCTTTAGA

2101 GATCGTTCA AGATTGCAGC TTGTGATGCA AGTTTCTCC  
AGCCAGAAAA

2151 CCTCATTTA AACCATCTGC TACTGGTAAT TCATACCAAT  
GCATTTCTT

2201 GGTGCTCGAT TTACACTATA ACCAAAGTTA AGTATTACAT  
TCAGGTGCTA

2251 CAACTTTCTA ATTTACAACC GAAACAAACA AGCAAACAGC  
ACTTGCTTG

2301 CTAATAACCC CATGGTGTAT TTTCCFTTT TATGATGACA  
AAACCAAGTA

2351 CATATGGTTT TATGTAGCAT TCAATTATAC TTCAGTGCTA  
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2401 ATGTTATAAG CAATTTGTAT TAAATCAGT TTTCCTTGAG  
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TGAACAGGAA

2501 TGTATCTTT ATTAGTATTG TTAATTGTGT TACTAATACT  
ATGCATATGA

2551 ATGAGAGCAA TGTATTCTA GGAGAACTCA GATATACATT  
CAACAATTTC

2601 TGTAGGTGAA AATGCATTAA CTGATGAAAG TTGAATCGTT  
AATGAGGGAG

2651 AAAACTGGGT ATCCATCCAT CCAACTATGT TAGGTGTTCA  
CCTGGTCTGT

2701 ATGTGACACC ACGCTGTTG GGTATCTCTC ACTTCACAT  
ACCTGTTCTC

2751 ATGGTTCTG CTACTCACTG TATTTGCAG GAGAGAAACA  
AAATGAAATC

2801 ACTGTCACCT ACTATGCC CATCACATAA GAACAATGGG  
GCTTTGGTGA

2851 CTTGTTCATG ATTACATAAG ATGTTGCAG CAGAGCAGCA  
ATAGAACCAA

2901 CACCATCCAC AGTTCTTGCT TGCTCTGTTA TGACTCCCTT  
TGCTGTCTTT

2951 ATGGTTGCA TGTATGAAGA ATACACTGCC TAATTCTAAT  
GTTAAAAAGT

3001 CACTGGGGTC AGATCTAGAG CTTAAGTAAG CAGTCTGGGG  
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3051 TTTATATGTT CCATAAAATG GAAATAAACCA CCTCCATAAT  
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3101 AAAAAAAAAA A

Figure 4  
No. 1

Seq. ID

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51 FDGLALLHSF TFKNGEVYYR SKYLRSDTYN CNIeanrivv  
SEFGTMAYPD

101 PCKNIFAKAF SYLSHTIPEF TDNCLINIMK TGDDYYATSE  
TNFIRKIDPQ

151 TLETLDKVVDY SKYVAVNLAT SHPHYDSAGN ILNMGTTSIVD  
KGRTKYVLFK

201 IPSSVPEKEK KKSCFKHLEV VCSIPSRSLL QPSYYHSFGI  
TENYIVFIEQ

251 PFKLDIVKLA TAYIRGVNWA SCLSFHKEDK TWFHFVDRKT  
KKEVSTKFYT

301 DALVLYHHIN AYEEDGHVVF DIVAYRDNSL YDMFYLKLLD  
KDFEVNNKLT

351 SIPTCKRFVV PLQYDKDAEV GSNLVKLPTS ATAVKEKDGS  
IYCQPEILCE

401 GIELPRVNYD YNGKKYKYVY ATEVQWSPVP TKIAKLNQQT  
KEVLHWGEDH

451 CWPSEPIFVP SPDAREEDEG VVLTCVVVSE PNKAPFLLIL  
DAKTFKELGR

501 ATVNVEMHLD LHGMFIPQND LGAETE

Figure 5

Seq ID No. 4 and Seq ID No. 5

437 NVQTKEVLH..WGEDHCWPSEPIFVPSPDAREEDEGVVLTCVVVSEPNKA  
484 ||.||| | | :||||||| ||| |||:|||.|| .  
465 NVKTKETWFTVWQEPDSYPSEPIFVSHPDALEEDDGVVLSVVSPGAGQK  
514  
485 P.FLLILDAKTFKELGRA.TVNVEMHLDLHGMF 515  
| :|||||.|||.||| || : . . ||:|  
515 PAYLLILNAKDLSEVARAEFTVEINIPVTFHGLF 548

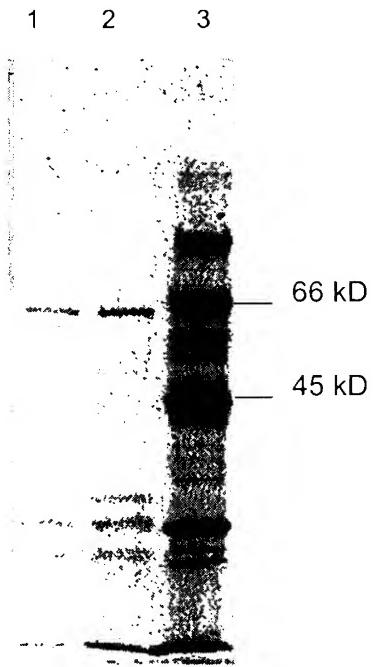


Fig. 6 shows a 10% polyacrylamide gel with E.coli expressed  $\beta,\beta$ -carotene 15,15'-monooxygenase after affinity tag purification; lane 1 and lane 2: 2 fractions from the  $\text{Co}^{2+}$ -chelate column showing the main band at 60 kD; lane 3: low range molecular weight marker (Bio Rad).

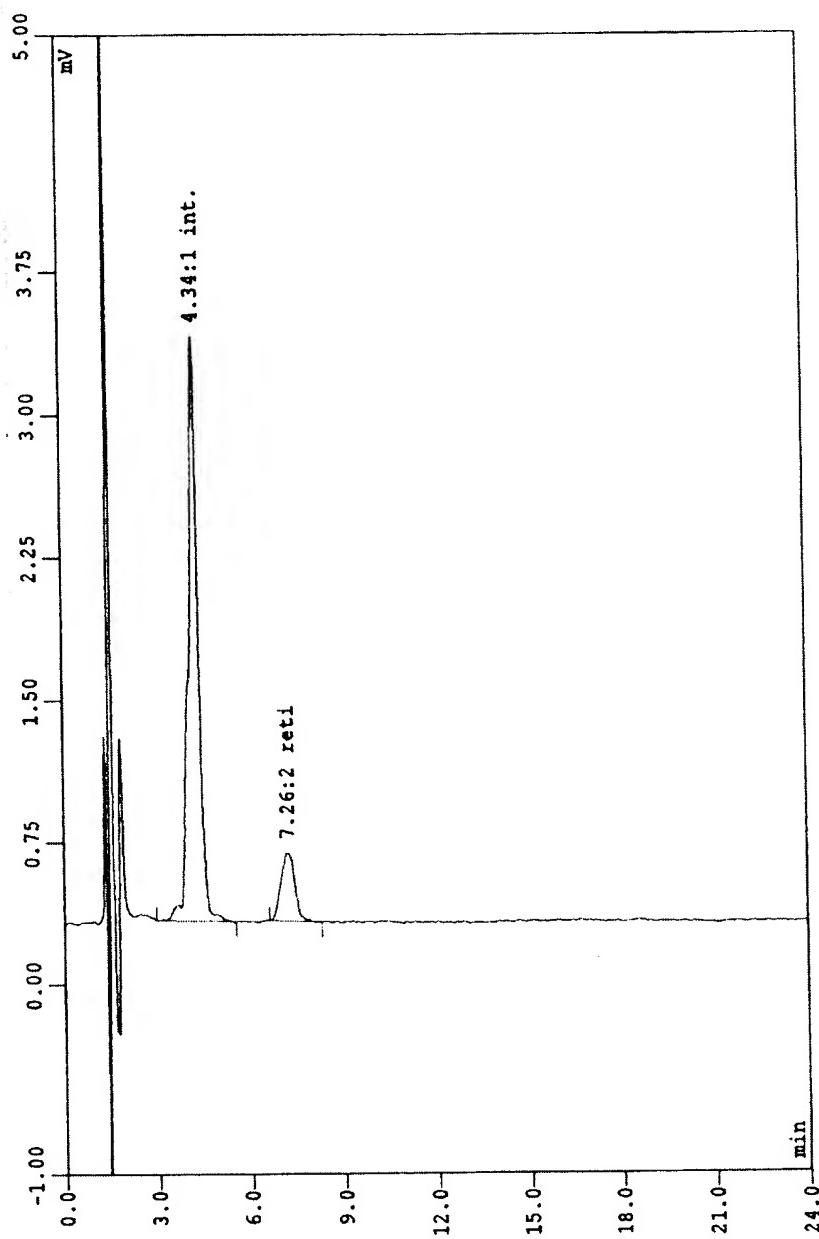


Fig. 7 shows an HPLC profile of the reaction mixture at the end of an activity assay for the  $\beta,\beta$ -carotene 15,15'-monooxygenase following the procedure in example 1. The first peak in the chromatogram represents the internal standard, while the second peak corresponds to retinal as the only product formed during the central cleavage with  $\beta$ -carotene as substrate.

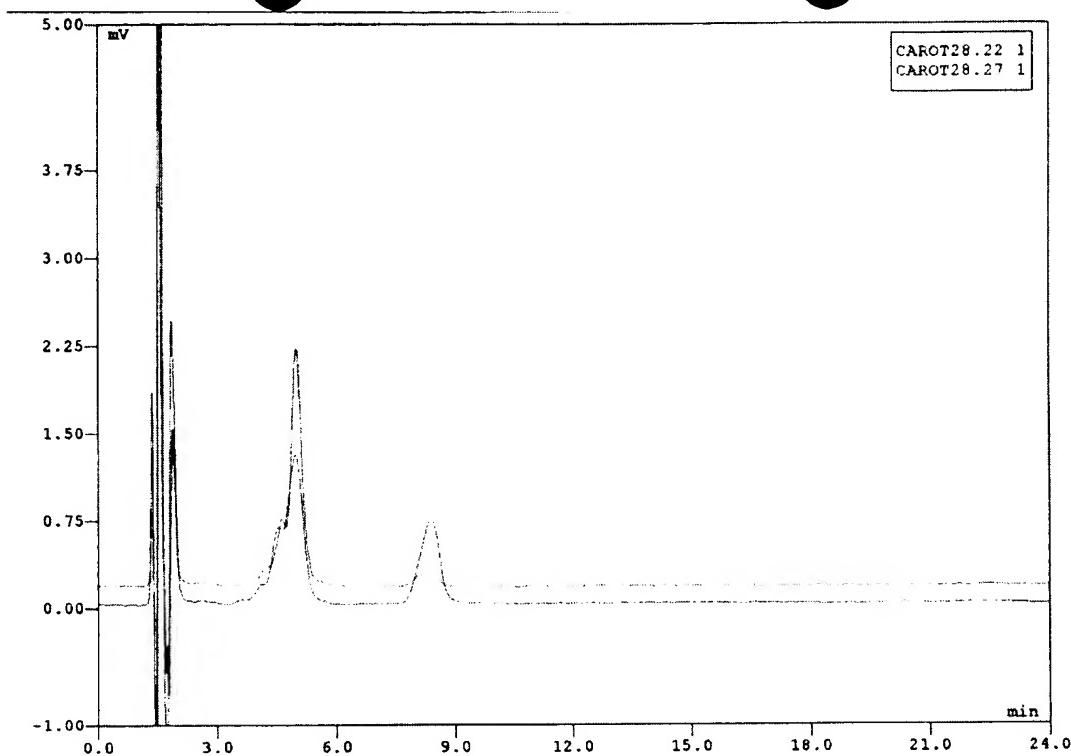


Fig. 8 confirms that the product peak in Fig. 7 is indeed retinal. A sample which was positive in the activity assay (green (upper) chromatogram) was spiked with retinal and analysed in second HPLC run (red (lower) chromatogram). The chromatograms of the two runs were then overlayed.